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Reactions of *Hordeum spontaneum* to infection with two cultures of *Puccinia hordei* from Israel and United States

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Summary

The reactions to infection with two cultures of *Puccinia hordei* were determined for 292 *Hordeum spontaneum* (syn. *H. vulgare* ssp. *spontaneum*) accessions, collected at 16 sites which encompassed the ecological range of *H. spontaneum* in Israel. Culture Tel-Aviv was from Israel and culture 57.19 was from the United States. Fifty-two percent of the accessions were resistant to culture Tel-Aviv and 67% were resistant to culture 57.19. Forty-three percent of the accessions were resistant to both cultures. The average infection type (IT) of accessions within sites ranged from 2.7 to 7.5 on a 0–9 rating scale. The results showed that the presence of *Ornithogalum* species, the alternate hosts of *P. hordei,* may increase the percentage of *H. spontaneum* accessions resistant to *P. hordei.* More accessions were resistant at sites where humidity at 1400 was higher, the annual evaporation was lower, and where the glumes were shorter. Kernel weight and annual rainfall was not correlated with resistance. A lower percentage of *H. spontaneum* accessions were resistant to *P. hordei* culture Tel-Aviv from Israel than to culture 57.19 from the United States. In a previous study a lower percentage of *H. spontaneum* accessions also was found to be resistant to a culture of *Erysiphe graminis hordei* from Israel than to cultures from other countries. Previous studies also have shown that cultures of *P. hordei* and *E. graminis hordei* from Israel have many genes for virulence on barley, and that *H. spontaneum* accessions from Israel have many genes for resistance to these two pathogens. Previous results and the results reported in this paper support the hypothesis of coevolution of resistant host genes and virulent pathogen genes where hosts and pathogens have coexisted for many thousand years.

Introduction

Collecting and evaluating *Hordeum spontaneum* (syn. *H. vulgare* L. ssp. *spontaneum* (C. Hock) Thell.) accessions obtained from plants growing in Israel has been receiving much attention. Seeds collected from several thousand *H. spontaneum* plants in Israel are being maintained in germplasm collections at several locations. The AFRC Institute of Plant Science Research at Cambridge, England, is maintaining seeds collected from over 51,000 *H. spontaneum* plants (Anishetty et al., 1982). The Lieberman Germplasm Bank for Cultivated Cereals, Tel-Aviv University, at Tel-Aviv ,

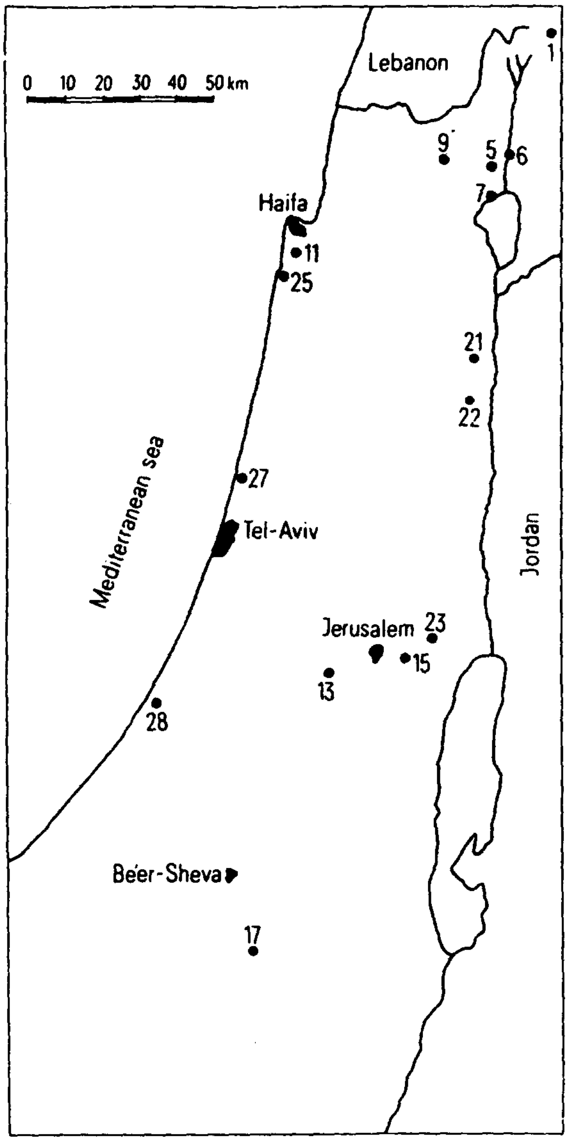

Fig. 1. Geographic distribution of 15 sites where *Hordeum spontaneum* was collected.

Israel, has over 20,000 accessions (personal communications I. Wahl). More than 3000 accessions are maintained in the USDA, ARS, National Small Grain Collection at Beltsville, MD, USA and in the Institute of Evolution, University of Haifa, at Haifa, Israel.

Many *H. spontaneum* accessions collected from plants in Israel have been found to be resistant to the barley leaf rust pathogen, *Puccinia hordei* Otth. Anikster et al. (1976) identified several *H. spontaneum* accessions outstanding for resistance to cultures of *P. hordei* from Israel. They found that many of the accessions collected in central or northern Israel near rust infected plants of *Ornithogalum* species, which are the alternate hosts for *P. hordei*, were resistant, whereas, few of the accessions collected in more arid regions were resistant. Moseman et al. (1980) reported that 49% of 290 *H. spontaneum* accessions collected in Israel were resistant to culture 57.19 of *P. hordei*. They also found that 41% of the accessions were resistant to culture 57.19 of *P. hordei* and to two cultures of the pathogen *Erysiphe graminis* DC. Fr. f. sp. *hordei* (Em. Marchal). In a continuation of previous studies, Nevo et al. (1979) reported that allozymic variations in proteins indicated that *H. spontaneum* accessions collected at 28 sites were genetically diverse, and that genetic diversity is correlated with environmental and ecological conditions. Moseman et al. (1983) found that *H. spontaneum* accessions collected at sites in the Galilee, Northern Plains and Judean Highlands were mostly resistant or moderately resistant to *E. graminis hordei* and that accessions collected in the arid Negev were mostly susceptible. Nevo et al. (1984) determined the correlations of allozymic variations, and ecological factors with resistance to *E. graminis hordei* in *H. spontaneum*. We are reporting the reactions of 292 *H. spontaneum* accessions collected at 16 sites in Israel to two cultures of *P. hordei*, one from the United States, 57.19, and one from Israel (Tel Aviv).

Materials and methods

The 292 accessions used in this study, which were collected at 16 sites in Israel (see Nevo et al., 1979 for sampling method) has been used in pevious studies (Moseman et al., 1983; Nevo et al., 1984). The sites, shown in Fig. 1, were chosen to cover the ecological and geographic range in which *H. spontaneum* grows in natural conditions in Israel.

The two cultures of *P. hordei*, 57.19 and Tel-Aviv, were chosen to represent the pathogenicity of cultures present in the United States and in Israel. Culture 57.19 of race 4 has been used in studies in which genes for resistance of the host have been identified (Roane & Starling, 1967). Culture Tel-Aviv, furnished by M. Reinhold, was isolated from a plant growing at Tel-Aviv, Israel (Reinhold & Sharp, 1982). Cultures 57.19 and Tel-Aviv have avirulence/virulence formulas Pal, 2, 2+, 2+ 5, 2+ 6, 3, 4, 7, 9/Pa8; and Pa2, 2+ 5, 2+ 6, 3, 7/Pal, 2+, 4, 8, 9, respectively.

The method of inoculation and development of infection was as follows: Plants in the first leaf stage were placed in an inoculation chamber and sprayed with a light weight oil to reduce surface tension. Urediospores, diluted with talcum powder, were gently blown on the plants with an atomizer. The inoculated plants were kept in moist inoculation chambers for about 16 hours at 16–20° C, and then dried for 2–4 hours under cool white fluorescent light at 24° C. The plants were maintained in a temperature and light controlled room in which the temperature was maintained at 16–19° C and illumination was provided with 175 UE $sec^{-1}m^{-2}$ of cool, white fluorescent light for 12 hours per day.

The reactions to infection were read nine days after inoculation and verified after 11 days. The infection types (ITs) produced by the reactions were read on a 0 to 9 scale from immune to susceptible as reported for reactions to infections with *E. graminis hordei* (Moseman et al., 1983). The reactions to infection were summarized by combining the ITs into 3 groups with resistant (R) = 0–3, moderately resistant (M) = 4–6, and susceptible (S) = 7–9. The average ITs for accessions from a site was the sum of the ITs on each accession divided by the number of accessions tested.

Table 1. Variations in reactions of *Hordeum spontaneum* accessions collected from 16 sites in Israel to infection with two cultures of *Puccinia hordei*

Location	Site no.	Number of accessions	Reactions to cultures 57.19 and Tel Aviv+									Average infection type
			RR	RM	MR	MM	RS	SR	MS	SM	SS	
			No. of accessions									
Damon	11	9	8	1								2.7
Mehola	22	20	11	7			1	1				3.0
Atlit	25	27	18	5	1		1		1		1	3.3
Herzliyya	27	21	11	3	3			2		2		3.3
Mt. Hermon	1	22	14	2	4		2					3.3
Mt. Meron	9	18	4	12		2						3.4
Ashqelon	28	28	13	6	2	1	3			3		3.5
Bet Shean	21	14	6	5		1			2			3.5
Bar Giyyora	13	14	8	1		2	1		1		1	3.6
Bor Mashash	17	11	5	2		1	3					3.6
Gadot	6	21	10	3	2	3	1		1	1		3.7
Rosh Pinna	5	16	9		1	2	1	2		1		3.8
Eyzariya	15	21	4		2	6	2		3		4	5.1
Tabigha	7	26	4	1	4	2	7		3	1	4	5.2
Wadi Qilt	23	12		1		2		2	1	3	3	5.9
Jericho Plains		12					1		2		9	7.5
Total	16	292	125	49	19	22	23	7	14	11	22	
%			42.8	16.8	6.5	7.5	7.9	2.4	4.8	3.8	7.5	3.96

+ Letters indicate reactions R(resistant) = IT's 0–3; M(moderately resistant) = IT's 4–6; and S(susceptible) = IT's 7–9. First letter indicates reactions to culture 57.19 and second letter indicates reactions to culture Tel-Aviv. For example: RM = Resistant to culture 57.19 and moderately resistant to culture Tel Aviv.

Table 2. Relationships of average infection types to annual rainfall, annual humidity at 1400, annual evaporation, glume length and kernel weight

Location	Site no.	Average infection type	Rainfall annual (mm)	Humidity at 1400 annual (mean)	Evaporation annual (cm)	Glume length mean (mm)	Kernel weight (mg)
Damon	11	2.7	686	58.8	190	18.7	35.9
Mehola	22	3.0	290	37.5	240	13.4	19.6
Atlit	25	3.3	563	57.5	180	21.4	46.8
Herzliyya	27	3.3	521	63.4	180	17.8	30.8
Mt. Hermon	1	3.3	1400	48	–	19.1	31.9
Mt. Meron	9	3.4	1010	47.5	260	16.3	29.8
Ashqelon	28	3.5	424	64.0	190	14.2	34.1
Bet Shean	21	3.5	290	43.7	239	14.1	14.5
Bar Giyyora	13	3.6	537	47.1	215	15.8	39.4
Bor Mashash	17	3.6	108	43.3	260	21.0	26.6
Gadot	6	3.7	451	43.6	220	–	–
Rosh Pinna	5	3.8	697	43.6	220	17.2	41.8
Eyzariya	15	5.1	380	42.5	240	25.5	26.1
Tabigha	7	5.2	436	42.2	225	26.1	45.8
Wadi Qilt	23	5.9	144	34.7	230	24.9	26.1
Mean		3.8	529	47.8	221	19.0	32.1
Std. Dev.		0.89	331	09.0	27	04.3	09.4

[+] Data on annual rainfal, humidity at 1400, evaporation, glume length and kernel weight from Nevo et al. 1979.

Results

The reactions of the 292 accessions to infection with cultures 57.19 and Tel-Aviv are shown in Table 1. Forty-three percent of the accessions were resistant and 74% of the accessions were resistant or moderately resistant to both cultures. The average ITs of the accessions within sites ranged from 2.7 in Damon (site 11) to 7.5 in Jericho Plains. The average ITs in Eyzariya (site 15), Tabigha (site 7), Wadi Qilt (site 23) were 5.1, 5.2, 5.9, respectively. The average infection types in the other 11 sites ranged from only 3.0 to 3.8.

The relationships of average infection types to the ecogeographic conditions annual rainfall, humidity at 1400 hr and annual evaporation and the morphological characteristics glume length and kernel weight are shown in Table 2. The annual rainfall was highly variable ranging from 108 mm at Bor Mashash to 1400 at Mt. Hermon. The standard

Table 3. Reactions of 292 *Hordeum spontaneum* accessions from Israel to infection with cultures of *Puccinia hordei* from Israel and the United States

Country	Culture	Reaction						Total
		Resistant		Moderate		Susceptible		
		No.	%	No.	%	No.	%	
Israel	Tel Aviv	151	52	82	28	59	20	292
U.S.A.	57.19	197	67	55	19	40	14	292

Table 4. Plant inventory (PI) numbers of 124 *Hordeum spontaneum* accessions collected in Israel resistant to 2 cultures of *Puccinia hordei* maintained in USDA, ARS, National Small Grains Collection at Beltsville, MD

PI No.						
466250	466251	466252	466255	466256	466258	466259
466260	466264	466265	466266	466268	466269	466271
466272	466274	466275	466276	466277	466278	466279
466280	466284	466285	466293	466305	466310	466311
466312	466313	466321	466322	466323	466324	466325
466328	466329	466332	466333	466334	466335	
466336	466337	466338	466339	466341	466342	466344
466346	466348	466351	466353	466356	466360	466362
466363	466366	466368	466372	466373	466375	466394
466409	466410	466413	466427	466430	466431	466432
466434	466436	466437	466442	466443	466444	466445
466448	466450	466451	466453	466454	466456	466458
466459	466461	466470	466473	466476	466477	466479
466480	466481	466482	466483	466486	466492	466496
466505	466510	466513	466514	466516	466525	466526
466528	466532	466536	466537	466538	466539	466540
466541	466542	466566	466567	466568	466572	466573
466574	466575	466576	466578	466581	466584	

deviation in comparison with the mean was much higher for annual rainfall than for the other ecogeographic conditions or the morphology characteristics. This may explain why the positive correlation between resistance (= low infection type) and annual rainfall was low and nonsignificant ($r = 0.38$; $p = 0.08$). The average resistance was highly negatively correlated with glume length ($r = -0.76$; $p = 0.001$) and positively correlated with humidity at 1400 hr ($r = 0.54$; $p = 0.02$) and negatively with evaporation ($r = -0.28$; $p = 0.16$). Average infection types were independent of kernel weight with the correlation of $r = 0.03$.

The reactions of the 292 accessions to cultures 57.19 from the United States and Tel-Aviv from Israel are compared in Table 3. Fewer accessions were resistant to culture Tel-Aviv than to culture 57.19. There were 52% of the accessions resistant to culture Tel-Aviv and 67% of the accessions resistant to culture 57.19.

Correlation coefficients were determined between reactions of 286 *H. spontaneum* accessions to cultures 57.19 and Tel-Aviv of *P. hordei* and a composite of cultures 59.11 and R1 and culture 64.54 of *E. graminis hordei* included in a previous study (Moseman et al., 1983). The correlations between the reactions to the two cultures of *P. hordei* and between the composite of cultures and the single culture of *E. graminis hordei* were 0.499 and 0.442, respectively, which is significant at the

Table 5. Plant inventory (PI) numbers of 40 *Hordeum spontaneum* accessions collected in Israel resistant to three cultures of *Erysiphe graminis hordei* and to two cultures of *Puccinia hordei* maintained in USDA, ARS, National Small Grains Collection at Beltsville, MD

PI No.						
466272	466274	466275	466276	466277	466279	466293
466321	466322	466323	466324	466325	466328	466329
466332	466333	466334	466335	466336	466337	466338
466341	466344	466351	466409	466448	466450	466451
466453	466456	466458	466461	466492	466496	466505
466510	466532	466572	466574	466575		

0.001 level. The correlations between the two cultures of *P. hordei* and the three cultures of *E. graminis hordei* range from -0.10 to $+0.10$, which is not significant.

The 125 accessions resistant to cultures 57.19 and Tel-Aviv of *P. hordei* are listed in Table 4. The 40 accessions resistant to cultures 57.19 and Tel-Aviv of *P. hordei* and to cultures 59.11, R1 and 64.54 of *E. graminis hordei* are listed in Table 5.

Discussions and conclusions

The frequency and distribution of *H. spontaneum* plants growing in Israel, which are resistant to *P. hordei,* agrees with a previous study (Moseman et al., 1980). In the present study 43% of the plants were resistant and in the previous study 49% of the plants were resistant. Anikster et al. (1976) reported that more plants resistant to *P. hordei* were found in Central and Northern Israel, where plants of *Ornithogalum* species are often found to be infected with *P. hordei* than in arid regions where *P. hordei* is seldom observed. In the present study many resistant plants were found in the Coastal Plains, where *Ornithogalum* species are present, and the annual rainfall and humidity are high and annual evaporation is low, and fewer resistant plants were found in regions east of Jerusalem and in the Negev where the annual rainfall and humidity are low and the annual evaporation is high.

The relationships between the resistance of *H. spontaneum* accessions to cultures of *P. hordei* isolated in Israel and in United States were similar to the resistance of *H. spontaneum* to cultures of *E. graminis hordei* isolated in Israel and in other countries (Moseman et al., 1983). The percentage of *H. spontaneum* accessions resistant to cultures from Israel of both pathogens, was less than the percentage of accessions resistant to cultures from other countries. In this study 52% of the accessions were resistant to culture Tel-Aviv from Israel and 67% of the accessions were resistant to culture 57.19 from the United States. In a previous study (Moseman et al., 1983) with *E. graminis hordei,* 42% of the accessions were resistant to culture 64.54 from Israel and 59% of accessions were resistant to a composite of two cultures, one from the United States and the other from Japan.

The lower percentage of *H. spontaneum* accessions which were resistant to culture Tel-Aviv from Israel than to culture 57.19 from the United States indicates that fewer genes for resistance in the host were effective against the culture of the pathogen from Israel. This supports the hypothesis that resistance genes in the host and virulence genes in the pathogen coevolve in areas where the host and the pathogen have coexisted for many thousands of years and that fewer resistance genes are effective in the hosts against the virulence genes in the pathogens from the areas where they have coevolved.

The *H. spontaneum* accessions identified as resistant to *E. graminis hordei* and to *P. hordei* may increase and stabilize barley production in United States and worldwide. The Barley Working Group for the European Cooperative Programme for Conservation and Exchange of Crop Genetic Resources (ECP/GR) lists as first priority the procurement of data on resistance to those two pathogens. The resistance to those pathogens is inherited (one to four genes), conditions slow powdery mildewing and leaf rusting and are easily transferred to cultivated barley. The resistance genes are being conserved and recombined. More than one hunderd of the resistant accessions have been crossed with genetic male sterile cultivars to develop a composite cross population and seed of all the accessions had been increased and the accessions have been included in the USDA, ARS, National Barley Collection at Beltsville, where they are available to all breeders.

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